JOURNAL OF Neuroscience Research

A role for T cell exhaustion in Long COVID-19 and severe outcomes for several categories of COVID-19 patients

Roe, Kevin

Review timeline:

Submission date: 16 February 2021
Editorial Decision: Conditional Reject (RER) (2 March 2021)
Revision Received: 3 March 2021
Editorial Decision: Major Modification (19 April 2021)
Revision Received: 27 April 2021
Editorial Decision: Minor Modification (31 May 2021)
Revision Received: 1 June 2021
Accepted: 8 June 2021

Editor 1: Karina Alvina
Editor 2: Cristina Ghiani
Reviewer 1: Karina Alvina

1st Editorial Decision

Decision letter
Dear Dr Roe:

Thank you for submitting your manuscript to the Journal of Neuroscience Research, which has been evaluated by two editors. The comments are appended below. As you will see, we found the topic to be of potential interest, yet, the manuscript is not suitable for publication in its current form.

If you feel that you can adequately address the concerns of the reviewers, you may revise and resubmit your paper within 90 days. It will require further review. Please explain in your cover letter how you have changed the present version. If you require longer than 90 days to make the revisions, please contact Dr Cristina Ghiani (cghiani@mednet.ucla.edu). To submit your revised manuscript: Log in by clicking on the link below https://wiley.atyponrex.com/submissionBoard/1/d953820c-5751-41f8-a7a8-37274e512c28/current

(If the above link space is blank, it is because you submitted your original manuscript through our old submission site. Therefore, to return your revision, please go to our new submission site here (submission.wiley.com/jnr) and submit your revision as a new manuscript; answer yes to the question "Are you returning a revision for a manuscript originally submitted to our former submission site (ScholarOne Manuscripts)? If you indicate yes, please enter your original manuscript's Manuscript ID number in the space below" and including your original submission's Manuscript ID number (jnr-2021-Feb-9560) where indicated. This will help us to link your revision to your original submission.)

Thank you again for your submission to the Journal of Neuroscience Research; we look forward to reading your revised manuscript.

Best Wishes,

Dr Karina Alvina
Associate Editor, Journal of Neuroscience Research

Dr Cristina Ghiani
Editor-in-Chief, Journal of Neuroscience Research

Associate Editor: Alvina, Karina

JOURNAL OF Neuroscience Research

Comments to the Author:
The submitted manuscript "An explanation for the higher mortality rate of COVID-19 patients having schizophrenia, the higher mortality of patients in other categories and for Long COVID-19" by Dr Roe offers a possible hypothesis on how an existing parasitic infection that is associated with psychiatric conditions (such as schizophrenia) could explain the elevated death rate in certain cases of COVID and also certain symptoms that persist ("long covid").

While the idea is interesting and potentially demonstrable, this manuscript does not provide sufficient evidence to convince the reader that these phenomena are causally related, not just correlated. The manuscript also reads disjointed, and requires the reader to search for the main hypothesis. Further, while there are a few references cited to support these claims, are there any specific studies showing evidence of the link between parasitic infections, covid and psychiatric/long standing illness? even in this case, the evidence would be correlative not proof of causality. While this paper states a hypothesis (thus no data is required) more evidence is needed to fully accept that these correlations could be predictive of severity and/or duration of the disease.
For acceptance this manuscript could be reformatted so that the main claims are easily laid out, and supporting evidence is also easy to understand and accept.

Associate Editor: Ghiani, Cristina
Comments to the Author:

The letter to the editor focuses on an interesting topic, the possible link between preexisting infections and worse outcomes in certain COVID 19 patients with underlying psychiatric conditions, however, I feel that a through and extensive reorganization is needed, as the author does not make clear such link until quite late in the letter: (Therefore, SARS-CoV-2 viral interactions with the neuroinflammation, neurotransmitter disruptions and immune system dysfunctions caused by latent protozoan parasite infections in the human brain and central nervous system, even in milder latent infections that do not result in schizophrenia, are likely to cause very significant neurological and psychological symptoms. A latent protozoan parasite infection in the brain or central nervous system, such as by protozoan parasites including T. gondii, can harm the host immune system and facilitate SARS-CoV-2 viral entry into the brain to create long-term neurological and psychological symptoms in the brain and central nervous system, and this interaction could make a major contribution to many of the observed severe symptoms of COVID-19 or Long COVID-19.8-10).
He may want to consider beginning by with the hypothesis and supporting arguments, the covid Neuroscience symptoms, risks.

The first two paragraphs seems completely unrelated and disconnected from the rest of the commentary and its scope.

Some sentences need to be reworded, few examples:
"CD8 T cell exhaustion has also been observed in T. gondii infected humans, which is also hypothesized to cause T. gondii reactivations and localized inflammations with very severe consequences.3 T cell exhaustion is very harmful, because CD8 T cell exhaustion and a reduced host antiviral response have also been observed in COVID-19 patients who develop more severe infections and outcomes.4-5"
this can be simplified.

This sentence is confusing, why would mortality be " defined as death or discharge to hospice within 45 days of a positive SARS-CoV-2 test result.'?

the inflammation cytokine interleukin-6 is elevated in .....

Authors' Response

1

Response to the Reviewers' comments on the manuscript entitled *"An explanation for the higher mortality rate of COVID-19 patients having schizophrenia, the higher mortality rates of patients in other categories and for Long COVID-19"* submitted to the Journal of Neuroscience Research - Manuscript # jnr-2021-Feb-9560

Thank you very much for giving me the opportunity to amend the paper in response

to the reviewers comments.

Response to All Comments - The reviewers have clearly spent time to read the paper, and these are very helpful comments, which are both valuable and sincerely appreciated. I have revised the paper to address the reviewers' comments and also have made wording revisions to improve the paper's clarity. I have indicated new text by red coloring. If I have missed anything, please let me know. I will address any problem.

Each Review Comment is listed in bold italics. The Responses and the Revisions are included just below each Reviewer Comment in RED. Revisions in paper are summarized below.

Associate Editor: Alvina, Karina Comments to the Author: The submitted manuscript "An explanation for the higher mortality rate of COVID-19 patients having schizophrenia, the higher mortality of patients in other categories and for Long COVID-19" by Dr Roe offers a possible hypothesis on how an existing parasitic infection that is associated with psychiatric conditions (such as schizophrenia) could explain the elevated death rate in certain cases of COVID and also certain symptoms that persist ("long covid"). While the idea is interesting and potentially demonstrable, this manuscript does not provide sufficient evidence to convince the reader that these phenomena are causally related, not just correlated.

Response to the comment -

More persuasive evidence is now included. Four new references are added. The text paragraphs have been extensively rearranged and several references have been rearranged in accordance with the reorganization of the text. And new text has been added for clarification and is marked in red color - some samples are listed below -

Latent protozoan parasite infections, such as infections by *T. gondii*, can not only explain the increased mortality rate for schizophrenic COVID-19 patients, but these protozoan parasite infections can also be a major contributing factor and explanation for patients suffering unexpected severe COVID-19 symptoms or patients suffering what is now called "Long COVID-19" or "Long-COVID," with formerly infected patients called "COVID long-haulers."[1-3] A very significant fraction of patients who apparently ended their COVID-19 viral infections, regardless of their severity or mildness, still suffer a number of neurological and psychological symptoms of Long COVID-19 for several

months or longer, which include: fatigue, dyspnoea (shortness of breath), chest pain, joint pain, anosmia (impaired sense of smell), dysgeusia (impaired sense of taste), hair loss, cognitive problems including memory and attention deficits and psychological problems, such as anxiety, depression and sleep disorders. [1-3]

Considering the numerous and diverse symptoms of Long COVID-19, it is noteworthy that virtually all of these symptoms closely match the symptoms of toxoplasmosis from activated or reactivated *T. gondii*.[15-17] Patients with toxoplasmosis have unifocal or multifocal brain lesions and can suffer a large selection of intracerebral and extracerebral symptoms, including dyspnoea, fevers, seizures, headaches, changes in vision, altered mental status, focal neurological deficits, mental confusion, cognitive dysfunction, ataxia, various cranial nerve palsies selectively affecting the 12 cranial nerve functions (e.g., cranial nerve I palsy causes anosmia, cranial nerve VII, IX, and X palsy causes dysgeusia, etc.), behavioral or psychomotor changes, involuntary movements, pneumonia

and chorioretinitis.[15-17]

The manuscript also reads disjointed, and requires the reader to search for the main hypothesis.

Response to the comment -

A new introductory paragraph is added to explain the mysterious issues with mortality rates and symptoms, and provide the main hypothesis that explains how multiple pathogen infections could work together to explain virtually all the mysteries in mortality and symptoms of COVID-19 and Long COVID-19 -

There have been several mysterious and strange subpopulation mortality rate differences and symptoms associated with both COVID-19 and the collection of long duration postinfection symptoms known as Long COVID-19.[1-7] Some patients in various age and health categories have been virtually asymptomatic, some patients in the same categories have suffered several strange symptoms, and yet other patients in the same categories have died within a few days after viral infection by SARS-CoV-2.[1-7] It is hereby hypothesized that these mysterious and strange subpopulation mortality rate differences and symptoms are the result of a SARS-CoV-2 virus infection working in a bidirectional synergy with one or more existing latent pathogen infections in certain patients, through

induced mutually beneficial immune cell dysfunctions, including T cell exhaustion, etc.[6-10] For several reasons that will be discussed in detail later, the most likely latent pathogen infection involved is the very pervasive protozoan parasite *Toxoplasma gondii*.[8-10]

Further, while there are a few references cited to support these claims, are there any specific studies showing evidence of the link between parasitic infections, covid and psychiatric/long standing illness? even in this case, the evidence would be correlative not proof of causality.

Response to the comment -

So far as I can determine, there are no studies to link latent pathogen infections to the mysterious mortality rates and symptoms of either COVID-19 or Long COVID-19. This is an entirely new breakthrough explanation that can logically explain virtually all the observed mysteries in symptoms and subpopulation mortality rates of COVID-19 and Long COVID-19. This has probably been missed for several reasons, including the low likelihood of *T. gondii* protozoan parasite infections in the brain and CNS tissues of murine laboratory animals used for COVID-19 studies and experiments.

The hypothesis of this letter meets the criteria of Occam's razor - the simplest hypothesis with the fewest assumptions is usually the right explanation.

While this paper states a hypothesis (thus no data is required) more evidence is needed to fully accept that these correlations could be predictive of severity and/or duration of the disease. For acceptance this manuscript could be reformatted so that the main claims are easily laid out, and supporting evidence is also easy to understand and accept.

Response to the comment -

New paragraphs have been added. More evidence regarding the very close match in symptoms between toxoplasmosis (i.e., an active or reactivated *T. gondii* infection) and the symptoms of Long COVID-19 is also now included. Some samples are listed below -

There have been several mysterious and strange subpopulation mortality rate differences and symptoms associated with both COVID-19 and the collection of long duration postinfection symptoms known as Long COVID-19.[1-7] Some patients in various age and

health categories have been virtually asymptomatic, some patients in the same categories have suffered several strange symptoms, and yet other patients in the same categories have died within a few days after viral infection by SARS-CoV-2.[1-7] It is hereby hypothesized that these mysterious and strange subpopulation mortality rate differences and symptoms are the result of a SARS-CoV-2 virus infection working in a bidirectional
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synergy with one or more existing latent pathogen infections in certain patients, through induced mutually beneficial immune cell dysfunctions, including T cell exhaustion, etc.[6-10] For several reasons that will be discussed in detail later, the most likely latent pathogen infection involved is the very pervasive protozoan parasite *Toxoplasma gondii*.[8-10]
.....
Therefore, latent protozoan parasite infections, such as infections by *T. gondii*, can not only explain the increased mortality rate for schizophrenic COVID-19 patients, and the increased mortality rates of other certain categories of COVID-19 patients. Furthermore, but these protozoan parasite infections can also be a major contributing factor and explanation for patients suffering unexpected severe COVID-19 symptoms or patients suffering what is now called "Long COVID-19" or "Long-COVID," with formerly infected patients called "COVID long-haulers."[1-3] A very significant fraction of patients who apparently ended their COVID-19 viral infections, regardless of their severity or mildness, still suffer a number of neurological and psychological symptoms of Long COVID-19 for several months or longer, which include: fatigue, dyspnoea (shortness of breath), chest pain, joint pain, anosmia (impaired sense of smell), dysgeusia (impaired sense of taste), hair loss, cognitive problems including memory and attention deficits and psychological problems, such as anxiety, depression and sleep disorders. [1-3]
Considering the numerous and diverse symptoms of Long COVID-19, it is noteworthy that these symptoms very closely match the symptoms of toxoplasmosis from activated or reactivated *T. gondii*.[15-17] Patients with toxoplasmosis have unifocal or multifocal brain lesions and can suffer a large selection of intracerebral and extracerebral symptoms, including dyspnoea, fevers, seizures, headaches, changes in vision, altered mental status, focal neurological deficits, mental confusion, cognitive dysfunction, ataxia, various cranial nerve palsies selectively affecting the 12 cranial nerve functions (e.g., cranial nerve I palsy causes anosmia, cranial nerve VII, IX, and X palsy causes dysgeusia, etc.), behavioral or psychomotor changes, involuntary movements, pneumonia and chorioretinitis.[15-17]
5
Associate Editor: Ghiani, Cristina Comments to the Author: The letter to the editor focuses on an interesting topic, the possible link between preexisting infections and worse outcomes in certain COVID 19 patients with underlying psychiatric conditions, however, I feel that a through and extensive reorganization is needed, as the author does not make clear such link until quite late in the letter: (Therefore, SARSCoV-2 viral interactions with the neuroinflammation, neurotransmitter disruptions and immune system dysfunctions caused by latent protozoan parasite infections in the human brain and central nervous system, even in milder latent infections that do not result in schizophrenia, are likely to cause very significant neurological and psychological symptoms. A latent protozoan parasite infection in the brain or central nervous system, such as by protozoan parasites including T. gondii, can harm the host immune system and facilitate SARS-CoV-2 viral entry into

the brain to create long-term neurological and psychological symptoms in the brain and central nervous system, and this interaction could make a major contribution to many of the observed severe symptoms of COVID-19 or Long COVID-19.8-10).He may want to consider beginning by with the hypothesis and supporting arguments, the covid Neuroscience symptoms, risks. The first two paragraphs seems completely unrelated and disconnected from the rest of the commentary and its scope.

Response to the comment -

A new introductory paragraph has been added to explain the mysteries, state the hypothesis and make a logical transition to flow into the paragraphs discussing protozoan parasite infections -

There have been several mysterious and strange subpopulation mortality rate differences and symptoms associated with both COVID-19 and the collection of long duration postinfection symptoms known as Long COVID-19.[1-7] Some patients in various age and health categories have been virtually asymptomatic, some patients in the same categories have suffered several strange symptoms, and yet other patients in the same categories have died within a few days after viral infection by SARS-CoV-2.[1-7] It is hereby hypothesized that these mysterious and strange subpopulation mortality rate differences and symptoms are the result of a SARS-CoV-2 virus infection working in a bidirectional synergy with one or more existing latent pathogen infections in certain patients, through induced mutually beneficial immune cell dysfunctions, including T cell exhaustion, etc.[6-10] For several reasons that will be discussed in detail later, the most likely latent pathogen infection involved is the very pervasive protozoan parasite *Toxoplasma gondii*.[8-10]

Some sentences need to be reworded, few examples:
"CD8 T cell exhaustion has also been observed in T. gondii infected humans, which is also hypothesized to cause T. gondii reactivations and localized inflammations with very severe consequences.3

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Response to the comment -

The text has been modified for clarification and is marked in red color, with deletions indicated by a strikethrough -

CD8 T cell exhaustion has also been observed in *T. gondii* infected humans, which is believed to ultimately induce also hypothesized to cause *T. gondii* reactivations and localized inflammations.[10]

T cell exhaustion is very harmful, because CD8 T cell exhaustion and a reduced host antiviral response have also been observed in COVID-19 patients who develop more severe infections and outcomes.4-5" this can be simplified.

Response to the comment -

The text has been modified for clarification and is marked in red color, with deletions indicated by a strikethrough -

T cell exhaustion, particularly CD8 T cell exhaustion, is very harmful, because CD8 T cell exhaustion and a reduced host antiviral response have also been observed in COVID-19 patients who develop more severe infections and outcomes.[6-7]

This sentence is confusing, why would mortality be " defined as death or discharge to hospice within 45 days of a positive SARS-CoV-2 test result.'? the inflammation cytokine interleukin-6 is elevated in .....

Response to the comment -

The paragraph has been modified for clarification and is marked in red color, with

deletions indicated by a strikethrough -

A strong link between schizophrenia and a higher mortality rate from SARS-CoV-2 has been recently observed, with an odds ratio of 2.67 compared to reference patients, after adjustment of the odds ratios for age, sex, race and medical risk factors.[4] Mortality was defined as death within 45 days of a positive SARS-CoV-2 test result.[4] The COVID-19 risk factors for schizophrenia are extensive, including a high prevalence of smoking, cardiovascular disease, diabetes, chronic respiratory disease, antipsychotic medication effects, disparities in health care, etc.[5] However, diagnoses for mood and anxiety disorders were not associated with higher mortality after an apparently correct adjustment for age, sex, race and medical risk factors, where mortality was defined as death or

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discharge to hospice within 45 days of a positive SARS-CoV-2 test result.[4] Some anxiety disorders could represent milder *T. gondii* infections that cause significantly less immune system dysfunction and neurotransmitter disruptions, and this could be one explanation why only schizophrenia, but not anxiety disorders, is associated with increased SARSCoV-2 mortality rates.

.....

Coronaviruses, probably including the SARS-CoV-2 virus, can infect the brain and CNS through either a blood circulation pathway by entering the blood-brain barrier (BBB), and/or through a neuronal pathway by sensory nerves (e.g., the olfactory nerve) or by motor nerve endings.[18] The neurobiology of COVID-19 is only partly understood at this time, but it is known that the inflammation cytokine interleukin-6 is elevated in the blood plasma of COVID-19 infected patients.[14-15] Furthermore, an increased BBB permeability has also been observed from elevated levels of interleukin-1β and TNF-α in severe COVID-19 patients, where these elevated inflammatory cytokines likely first originated from injured lung epithelial tissues or blood vessel endothelial tissues.[19] Increased BBB permeability will also assist brain entry by neurotropic viruses, a viral category that includes the SARS-CoV-2 virus.[19-20] Unfortunately, most brain tissue studies of the SARS-CoV-2 virus appear to rely mainly on laboratory-raised murine brain tissue (which is unlikely to be infected with protozoan parasites such as *T. gondii*).[19-20]

2nd Editorial Decision

Decision Letter

Dear Dr Roe:

We've now received the reviewer feedback and have appended those reviews below. They found the developed hypothesis interesting, but, also raised a series of concerns and made some suggestions on the organization of the manuscript. In light of the comments, your manuscript is not suitable for publication in the present form.

If you feel that you can adequately address the concerns of the reviewers, you may revise and resubmit your paper within 90 days. It will require further review. Please explain in your cover letter how you have changed the present version and submit a point by point response to the editors' and reviewers' comments. If you require longer than 90 days to make the revisions, please contact Dr Cristina Ghiani (cghiani@mednet.ucla.edu). To submit your revised manuscript: Log in by clicking on the link below https://wiley.atyponrex.com/submissionBoard/1/3cd23ac6-4c2b-4b27-b2d3-e81396c292b1/current

(If the above link space is blank, it is because you submitted your original manuscript through our old submission site. Therefore, to return your revision, please go to our new submission site here (submission.wiley.com/jnr) and submit your revision as a new manuscript; answer yes to the question "Are you returning a revision for a manuscript originally submitted to our former submission site (ScholarOne Manuscripts)? If you indicate yes, please enter your original manuscript's Manuscript ID number in the space

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below" and including your original submission's Manuscript ID number (jnr-2021-Feb-9560.R1) where indicated. This will help us to link your revision to your original submission.)

Thank you again for your submission to the Journal of Neuroscience Research; we look forward to reading your revised manuscript.

Best Wishes,

Dr Karina Alvina
Associate Editor, Journal of Neuroscience Research

Dr Cristina Ghiani
Editor-in-Chief, Journal of Neuroscience Research

Associate Editor: Alvina, Karina
Comments to the Author:
In this letter to the editor, The author posits the very interesting hypothesis that somehow the increased mortality in schizophrenic patients affected with covid-19 is related to pre-existing parasitic infection. While the idea that parasitic infections before covid could be the cause of higher death rate in certain individuals is interesting and provocative, it is highly correlational as there us no clear evidence of causality. Therefore, this manuscript requires extensive revision before it can be considered for acceptance.

Reviewer: 1

Comments to the Author
In this letter to the editor, Dr. Roe suggests that the higher mortality rate of COVID-19 patients having schizophrenia might be related to co-infection by the parasite Toxoplasma gondii, which shows predilection for brain tissue as a tachyzoite (acute infection) or bradyzoite (cyst) form. Although the idea is provocative, there is no causal evidence of T. gondii and SARS-CoV-2 co-infection in individuals with schizophrenia. A quick Pubmed search using the keywords COVID-19 and Toxoplasma provided 4 hits, all in cats, which are also highly susceptible to infections by other microbes including fungi. Similarly, the general neurological and psychological symptoms described by the author in lines 113-132, can be attributed to other brain pathogens such as the neurotropic Cryptococcus neoformans among others, which has been described in recent publications (PMID:33598258, 32596073, 33047097), not necessarily in schizophenic individuals but in those with T cell deficiency or immunosuppressed as described by the author in this piece. This fungus also causes destructive brain lesions in patients. In addition, whether the neurological and psychological symptoms described by the authors are present in individuals with COVID-19 and any secondary infection associated with the virus yet needs to be determined since the majority of these patients die before these manifestations can be documented. Furthermore, patients with T cell deficiency due to exhaustion as described by the author or in pre-existing immunosuppression rarely have a strong inflammatory response in the CNS and do not show considerable microglial activation. Finally, stronger evidence rather than correlative or coincidental information needs to be provided to support the current claims by the author.

Reviewer: 2

Comments to the Author
This brief paper provides an interesting hypothesis about some peculiar aspects of COVID-19 epidemiology and physiopathology, which so far haven't received any attention, despite the massive amount of publications on COVID-19 appeared in the last 12 months. I feel that this hypothesis is worth reading and may suggest a new avenue of research, so far missed. The author has well incorporated all the comments received so far. The paper is easy to read and might attract readers' attention.

I have one MAJOR comment: to facilitate the flow of ideas and facilitate reading, the author should add several HEADINGS to split the manuscript in different sections. Although the manuscript taps on different issues, so far the paper doesn't include any headings, and this does not help the reader contextualize the paper content.

Reviewer: 3

Comments to the Author
The paper develops an interesting topic,that currently has several quesions and few answers. The hypothesis pointed out by the authors is very interesting and deserves attention for future researches in the literature.
However I have some queries for the authors.
From line 54 to line 67 authors cite results from the review from Xiao et al., 2018 claiming that " these neurotransmitter disruptions have been hypothesized to cause schizophrenia" However the review exolains that the IgG levels could be also associated to the antipsychotic drug use, and authors should mention this in the text. Moreover the same review associates T.gondii IgG also to other psychiatric disorder and cognitive impairment. The authors should mention this in the text as well. At last, only 3/34 individuals with

schizophrenia were positive for the T.Gondii IgG, and this could explain the multifatoral risk factors for scizophrenia.
At page 4 (line 73) authors should mention also sedentary lifestyle as a risk factor for schizophrenia.
Still at page 4 (lines 90-92) authors affirm that "These higher
mortality rates closely match corresponding increases in latent pathogen
infection rates, particularly protozoan parasite infections, such as T. gondii. At the light of the current evidence authors should "soften" this sentence.
From lines 163-171 authors mention several other pathogen sinergies. In my opinion authors should move this paragraph at page 2 after line 37.
At the end of the paper, authors should make some reccomendations for researchers like suggesting a T.Gondii IgG research in patients with long COVID 19 or case control studies in patients with or without neurological or psychiatric symptoms following the infection.

Reviewer: 4

Comments to the Author
<pr>The submitted manuscript offers a hypothesis on how latent protozoan infections such as t. gondii may be responsible for differences in risk of death and risk of persistent symptoms due to Covid-19 in various populations. </p>
<pr><u>Latent infections as a risk factor for Covid-19 mortality</u>: The author describes increased prevalence of T. gondii antibodies found in patients with certain psychiatric conditions, in men (compared to women), those in older age groups, and adults with increased BMI--groups that are at increased risk of Covid-related mortality. He argues that latent parasitic infections may be responsible for increased risk in these populations. While this may be possible, what is the evidence for causation? It seems plausible that an underlying immune mechanism may predispose people to both Covid-19 and risk of other infections, but it's not clear based on the presented evidence that 1) this mechanism is unique to latent protozoan infections or that 2) latent protozoan infections cause the immune disruption that leads to poor Covid-19 related outcomes. Support for the first argument may come from studies that suggest that these risk factors (male sex, severe mental illness, increased BMI, older age) are unique to Covid-19 and T. Gondii, as opposed to risk factors for worse infectious outcomes in general. Support for the second argument could come from studies suggesting that latent protozoan infections have an adverse impact on outcomes in the setting of subsequent infections. Is there evidence for the "bidirectional synergy" or "mutually beneficial immune cell dysfunctions" referenced? What are these mechanisms, specifically? What is meant by "beneficial"? </p>
<pr><u>Latent infections as a risk factor for long COVID-19</u>: The author discusses the possibility that a variety of latent infections (viral, bacterial, protozoan) can act in synergy and contribute to symptoms of post-acute Covid-19 (long Covid). Is there evidence that the same risk factors for Covid-19 mortality referenced earlier in the paper are risk factors for long-Covid? The author describes how symptoms of long-Covid parallel symptoms of toxoplasmosis reactivation; how would you distinguish between the two? </p>
<pr><u>Summary</u>: This paper presents an interesting hypothesis concerning the possibility that latent parasitic infections contribute to adverse outcomes in the acute and post-acute phase of Covid-19 infection. Though shared immune mechanisms may contribute to increased risk of Covid-19 infection and other infections, it isn't clear based on the evidence presented that latent infections are a cause of adverse outcomes across various at-risk groups. Suggestions regarding how this hypothesis could be tested may help guide future research in this area.

Authors' Response

1

Response to the Reviewers' comments on the manuscript entitled *"An explanation for the higher mortality rate of COVID-19 patients having schizophrenia, the higher mortality rates of patients in other categories and for Long COVID-19"* submitted to the Journal of Neuroscience Research - Manuscript # jnr-2021-Feb-9560

Thank you very much for giving me the opportunity to amend the paper in response to the reviewers comments.

Response to All Comments - The reviewers have clearly spent time to read the paper, and these are very helpful comments, which are both valuable and sincerely appreciated. I have revised the paper to address the reviewers' comments and also have made wording revisions to improve the paper's clarity. I have indicated new text by red coloring. If I have missed anything, please let me know. I will address any problem.

Each Review Comment is listed in bold italics. The Responses and the Revisions are included just below each Reviewer Comment in RED. Revisions in paper are summarized below.

Associate Editor: Alvina, Karina Comments to the Author: In this letter to the editor, The author posits the very interesting hypothesis that somehow the increased mortality in schizophrenic patients affected with covid-19 is related to pre-existing parasitic infection. While the idea that parasitic infections before covid could be the cause of higher death rate in certain individuals is interesting and provocative, it is highly correlational as there us no clear evidence of causality. Therefore, this manuscript requires extensive revision before it can be considered for acceptance.

Some stronger evidence is now included in the revised paper with the addition of several new references.

Evidence of Microglial Activation and Microglial Nodules in Brain Autopsies of COVID-19 Fatalities

It has been recently reported that 41 patients, with ages ranging from 38 to 97 years, who died from COVID-19, had brain autopsies in 2020 at the Columbia University Irving Medical Center [11]. The patients tested before death showed elevated inflammatory cytokines, including elevated IL-6 in 26 (96%) of the 27 patients tested [11]. Neuropathological examination of 20 to 30 areas from each brain found microglial activation in 34 of the 41 brains (81%) [11]. Microglial clusters (microglial nodules) were found in 26 of the brains (63%), being most prevalent in the brainstem, and neurons were found in some of the microglial nodules, indicating neuronophagia [11]. However, viral RNA and viral proteins were not significantly detected in the brain cells or the microglial nodules, while acute and subacute hypoxic damage was seen in every brain; thus the formation of the microglial nodules was attributed to hypoxia, not SARS-CoV-2 [11].

It is well documented that brain hypoxia disrupts the BBB, causing leakage of proinflammatory plasma proteins, including immunoglobulins, fibrinogen and

complement [12]. It is also well documented that microglial activation and perivascular microglial clusters can be caused by fibrin deposition from perivascular leakage of plasma protein fibrinogen into areas of BBB disruption [13]. In addition, the formation of apoptotic neuron-astrocyte-microglia triads and extensive damage to the myelin sheath (demyelination) from oxidative stress and inflammatory stress has been well documented for brain hypoxia/ischemia [12]. But while neurons were found in some microglial nodules, no apoptotic neuron-astrocyte-microglia triads were reported, and there was absolutely no evidence of any demyelination seen in any of the brains autopsied [11]. If there was brain hypoxia, it was of such short duration that not even demyelination from the hypoxia death of oligodendrocytes could occur [12]. Therefore, brain hypoxia is probably not the main explanation for the microglial nodules. However, there is another very well-documented explanation for the microglial nodules that were found in these brain autopsies.

Extensive Evidence of Microglial Brain Nodules in *T. gondii* Infections

T. gondii infections have shown an ability to use microglia as "Trojan Horses" for intracellular tachyzoite replication and spreading these active protozoan parasites throughout the brains of infected hosts [14]. Infected microglia once activated have demonstrated an ability to transmigrate through astrocytes, which are part of the brain parenchyma and BBB [14]. Rapid transfers of these pathogens from infected glial cells to effector T cells has also been demonstrated [14]. Microglial nodules in the brain and CNS have also been widely observed in AIDS patients, where the microglial nodules were

caused by *T. gondii* infections, cytomegalovirus infections or HIV itself [14-16]. Activated microglia from reactivated *T. gondii* have also been observed to cause neuronal apoptosis, which could eventually lead to neuronophagia [16]. *T. gondii* activated microglia also lead to elevated levels of the inflammatory cytokines IL-1β, IL-6, and TNF-α [16].

In view of the preceding *T. gondii* symptoms, it is significant that microglial activation was also found in 81% of the brains of COVID-19 fatalities, and microglial nodules were found in 63% of the brains of COVID-19 fatalities [11]. But while neurons were found in some microglial nodules, no apoptotic neuron-astrocyte-microglia triads were reported, and there was absolutely no demylination found in any of the brains autopsied [11]. But one of the main consequences of brain hypoxia, BBB breakdown and oxidative and inflammatory stress is axonal demyelination of the neurons [12]. If there was brain hypoxia, it was of such short duration that not even demyelination occurred. Therefore, *T. gondii* infections are a more likely explanation for the microglial activation and microglial nodules found in the reported brain autopsies.

Conclusive evidence will only become available after a research effort requiring major resources in equipment and money. Publication of this paper is needed first in order to muster the resources in equipment and money needed to collect the necessary conclusive evidence.

Reviewer: 1 Comments to the Author In this letter to the editor, Dr. Roe suggests that the higher mortality rate of COVID-19 patients having schizophrenia might be related

to co-infection by the parasite Toxoplasma gondii, which shows predilection for brain tissue as a tachyzoite (acute infection) or bradyzoite (cyst) form. Although the idea is provocative, there is no causal evidence of T. gondii and SARS-CoV-2 co-infection in individuals with schizophrenia. A quick Pubmed search using the keywords COVID-19 and Toxoplasma provided 4 hits, all in cats, which are also highly susceptible to infections by other microbes including fungi. Similarly, the general neurological and psychological symptoms described by the author in lines 113-132, can be attributed to other brain pathogens such as the neurotropic Cryptococcus neoformans among others, which has been described in recent publications (PMID:33598258, 32596073, 33047097), not necessarily in schizophenic individuals but in those with T cell deficiency or immunosuppressed as described by the author in this piece. This fungus also causes destructive brain lesions in patients.

T. gondii is the strongest candidate and it is present in over two billion people world-wide (one out of three people have it). *Cryptococcus neoformans* prevalence is also high, in some countries 70% to 80% of all children over age five have antibodies to *C. neoformans*. It causes a million infections a year in HIV patients, so the prevalence of latent *C. neoformans* infections world-wide must be high. However, unlike *T. gondii*, *Cryptococcus neoformans* has never been repeatedly linked to schizophrenia (for any percentage of cases), or T cell exhaustion. But Other latent pathogens, such as *Cryptococcus neoformans*, are not, and never were, directly excluded from causing immune dysfunctions, such as T cell exhaustion. The main point is that these long duration latent pathogen infections can cause immune dysfunctions, such as T cell exhaustion, which would be very detrimental and lead to more severe COVID-19 infections, many becoming fatal. And COVID-19 infections can also cause more T cell

exhaustion that can partially or fully reactivate latent pathogen infections, especially the very widely prevalent *T. gondii*, but these reactivations are not being solely limited to *T. gondii*. Some new clarification is added.

Other Comorbidities And Pathogen Synergies With SARS-CoV-2 By T Cell Exhaustion

It should be noted that the mutually beneficial T cell exhaustion induced by a SARSCoV-2 virus infection and a latent pathogen infection is not exclusive to *T. gondii*. T cell exhaustion can possibly be caused by other latent infections of fungal or viral pathogens, including *Cryptococcus neoformans*, the hepatitis B virus, hepatitis C virus, and certain members of the alpha, beta and gamma herpes virus families, such as cytomegalovirus [32-37]. For example, a synergy between the SARS-CoV-2 virus and the hepatitis B and C viruses could explain the adjusted odds ratio of 2.62 in COVID-19 mortality that was observed in moderate to severe liver disease patients within a study of 31,461 COVID-19 patients [16]. In conclusion, several other latent pathogen infections that induce T cell exhaustion can also affect the outcome for COVID-19 patients, and explain the symptoms of patients with Long COVID-19 [1-3, 32-39].

Finally, there is a remarkably close match in the unusual and characteristic symptoms between Long COVID-19 and toxoplasmosis. This is discussed in the following modified paragraphs.

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It is noteworthy that the unusual symptoms of Long COVID-19 closely match the symptoms caused by a partially or fully reactivated *T. gondii* toxoplasmosis [21-25]. Toxoplasmosis patients also exhibit several matching symptoms, such as dyspnoea, fevers, seizures, headaches, changes in vision, altered mental status, focal neurological deficits, mental confusion, cognitive dysfunction, ataxia, behavioral or psychomotor changes, involuntary movements, pneumonia, chorioretinitis and a variety of cranial nerve palsies [20-26].

The most unusual and distinctive Long COVID-19 symptoms can be explained by toxoplasmosis induced cranial nerve palsies capable of affecting the functions of the 12 cranial nerves, that create symptoms such as: anosmia from a palsy of the olfactory nerve (I), vertigo (dizziness), tinnitus and otalgia (earache) from a palsy of the vestibulocochlear nerve (VIII), and dysgeusia from palsies of the facial nerve (VII), glossopharyngeal nerve (IX), and vagus nerve (X) [20-26]. Speech and swallowing deficits can be caused by nerve palsies of one or more of the following cranial nerves: trigeminal nerve (V), facial nerve (VII), glossopharyngeal nerve (IX), vagus nerve (X), accessory nerve (XI) and hypoglossal nerve (XII) [27].

Table 1 compares the symptoms of COVID-19 and/or Long COVID-19 and the matching symptoms possible from toxoplasmosis. One symptom discrepancy is chorioretinitis. Chorioretinitis has been sporadically reported in COVID-19 patients, but apparently not yet reported in Long COVID-19 patients [28].

In addition, whether the neurological and psychological symptoms described by the authors are present in individuals with COVID-19 and any secondary infection associated with the virus yet needs to be determined since the majority of these patients die before these manifestations can be documented.

Some stronger evidence is now included in the revised paper.

Evidence of Microglial Activation and Microglial Nodules in Brain Autopsies of

COVID-19 Fatalities

It has been recently reported that 41 patients, with ages ranging from 38 to 97 years, who died from COVID-19, had brain autopsies in 2020 at the Columbia University Irving Medical Center [11]. The patients tested before death showed elevated inflammatory cytokines, including elevated IL-6 in 26 (96%) of the 27 patients tested [11]. Neuropathological examination of 20 to 30 areas from each brain found microglial activation in 34 of the 41 brains (81%) [11]. Microglial clusters (microglial nodules) were found in 26 of the brains (63%), being most prevalent in the brainstem, and neurons were found in some of the microglial nodules, indicating neuronophagia [11]. However, viral RNA and viral proteins were not significantly detected in the brain cells or the microglial nodules, while acute and subacute hypoxic damage was seen in every brain; thus the formation of the microglial nodules was attributed to hypoxia, not SARS-CoV-2 [11].

It is well documented that brain hypoxia disrupts the BBB, causing leakage of proinflammatory plasma proteins, including immunoglobulins, fibrinogen and complement [12]. It is also well documented that microglial activation and perivascular microglial clusters can be caused by fibrin deposition from perivascular leakage of plasma protein fibrinogen into areas of BBB disruption [13]. In addition, the formation of apoptotic neuron-astrocyte-microglia triads and extensive damage to the myelin sheath (demyelination) from oxidative stress and inflammatory stress has been well documented for brain hypoxia/ischemia [12]. But while neurons were found in some microglial nodules, no apoptotic neuron-astrocyte-microglia triads were reported, and there was absolutely no evidence of any demyelination seen in any of the brains autopsied [11]. If there was brain hypoxia, it was of such short duration that not even demyelination from the hypoxia death of oligodendrocytes could occur [12]. Therefore, brain hypoxia is probably not the main explanation for the microglial nodules. However, there is another very well-documented explanation for the microglial nodules that were found in these brain autopsies.

Extensive Evidence of Microglial Brain Nodules in *T. gondii* Infections

T. gondii infections have shown an ability to use microglia as "Trojan Horses" for intracellular tachyzoite replication and spreading these active protozoan parasites throughout the brains of infected hosts [14]. Infected microglia once activated have demonstrated an ability to transmigrate through astrocytes, which are part of the brain parenchyma and BBB [14]. Rapid transfers of these pathogens from infected glial cells to effector T cells has also been demonstrated [14]. Microglial nodules in the brain and CNS have also been widely observed in AIDS patients, where the microglial nodules were caused by *T. gondii* infections, cytomegalovirus infections or HIV itself [14-16]. Activated microglia from reactivated *T. gondii* have also been observed to cause neuronal apoptosis, which could eventually lead to neuronophagia [16]. *T. gondii* activated microglia also lead to elevated levels of the inflammatory cytokines IL-1β, IL-6, and TNF-α [16].

In view of the preceding *T. gondii* symptoms, it is significant that microglial activation was also found in 81% of the brains of COVID-19 fatalities, and microglial nodules were found in 63% of the brains of COVID-19 fatalities [11]. But while neurons were found in some microglial nodules, no apoptotic neuron-astrocyte-microglia triads were reported, and there was absolutely no demylination found in any of the brains autopsied [11]. But

one of the main consequences of brain hypoxia, BBB breakdown and oxidative and inflammatory stress is axonal demyelination of the neurons [12]. If there was brain hypoxia, it was of such short duration that not even demyelination occurred. Therefore, *T. gondii* infections are a more likely explanation for the microglial activation and microglial nodules found in the reported brain autopsies.

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Furthermore, patients with T cell deficiency due to exhaustion as described by the author or in pre-existing immunosuppression rarely have a strong inflammatory response in the CNS and do not show considerable microglial activation.

T cell exhaustion is complicated. The discussion of T cell exhaustion has been greatly clarified, deepened and broadened in the revised paper. Some of the newly added paragraphs include the following -

There is a fundamental question concerning the prevalence and severity of T cell exhaustion. Why is T cell exhaustion so widely induced by both virulent and latent pathogen infections, and also induced by several cancers, such as leukemias and lymphomas [34-37]? T cell exhaustion is caused by persistent antigen exposures and persistent inflammation, and these conditions easily result from several long duration virulent pathogen infections (e.g., coronaviruses such as SARS-CoV-2), latent pathogen infections (e.g., protozoan parasites such as *T. gondii*) and cancers [34-35]. And the severity of T cell exhaustion is determined by both the abundance (titer) of the antigens, such as from increased pathogen numbers (e.g., increased pathogen and antigen levels from primary or secondary viremia), and by the time duration of the antigen stimulation [34-35]. This is significant, because this also strongly implies that longer duration latent pathogen infections and longer duration cancers will cause more severe T cell exhaustion. Therefore, older individuals will generally also be more likely to have severe T cell exhaustion with all its consequences for higher mortality. This would be one more logical explanation for the increasing mortality rate of older individuals.

T Cell Exhaustion Synergies Between Cancers and SARS-CoV-2

T cell exhaustion is associated with loss of T cell effector functions, reduced proliferative and cytotoxic capacities and impaired production of interleukin-2 and pro-inflammatory cytokines, and increased expression of inhibitory receptors, including the programmed cell death protein 1 (PD-1) receptor, cytotoxic T lymphocyte antigen-4 (CTLA-4) receptor, the lymphocyte-activation gene 3 (LAG3) receptor, and the T cell immunoglobulin domain and mucin domain 3 (Tim-3) receptor, etc. [26,34-35]. This is noteworthy because several cancers produce ligands, such as PD-L1 and PD-L2, that induce T cell exhaustion by binding to the inhibitory PD-1 T cell receptor [35]. Furthermore, several COVID-19 patients have certain cancer comorbidities that can cause T cell exhaustion and also have significantly increased odds ratios (O.R.) for critical illness and mortality, including lung cancers (O.R. of mortality 2.89), leukemias or lymphomas (O.R. of critical illness 3.53; O.R. of mortality 2.2), colorectal cancer (O.R. of mortality 1.73), etc. [11-12,35-40]. The worse outcomes for these COVID-19 cancer patients can be explained by a bidirectional synergy between the SARS-CoV-2

virus and certain cancers through T cell exhaustion and other immune cell dysfunctions.

Can Antigen-Specific T Cell Exhaustion Cause Other T Cells' Exhaustion?

One basic question that should be addressed is whether T cell exhaustion in antigenspecific T cells can affect other T cells that are not specific for the same antigen as the exhausted antigen-specific T cells. There are actually several reasons to believe that this can occur [34, 41-44]. T cell exhaustion can be caused by inhibitory receptors and

desensitization of co-stimulatory receptors for antigen-specific T cells [34]. However, there are also soluble mediators that inhibit T cell functions, including immunosuppressive cytokines interleukin-10 (IL-10), transforming growth factor-β (TGF-β), and indoleamine 2,3 dioxygenase (IDO); inflammatory cytokines including interferons α/β that can optionally also promote immune suppression of T cells; dendritic cells, macrophages and B cells that can be transformed into immunoregulatory antigen presenting cells that secrete elevated levels of IL-10, IDO, and TGF-β; upregulated immunoregulatory TREG cells; and myeloid-derived suppressor cells that can inhibit T cell functions and/or promote T cell exhaustion [34,41-44]. Therefore, there are multiple nonspecific pathways for antigen-specific T cell exhaustion induced by one pathogen or cancer to inhibit T cells and/or cause T cell exhaustion in naive T cells or other T cells specific for other antigens.

Finally, stronger evidence rather than correlative or coincidental information needs to be provided to support the current claims by the author.

Some stronger evidence is now included in the revised paper with the addition of several new paragraphs and references.

Evidence of Microglial Activation and Microglial Nodules in Brain Autopsies of COVID-19 Fatalities

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It is well documented that brain hypoxia disrupts the BBB, causing leakage of proinflammatory plasma proteins, including immunoglobulins, fibrinogen and complement [12]. It is also well documented that microglial activation and perivascular microglial clusters can be caused by fibrin deposition from perivascular leakage of plasma protein fibrinogen into areas of BBB disruption [13]. In addition, the formation of apoptotic neuron-astrocyte-microglia triads and extensive damage to the myelin sheath (demyelination) from oxidative stress and inflammatory stress has been well documented for brain hypoxia/ischemia [12]. But while neurons were found in some microglial nodules, no apoptotic neuron-astrocyte-microglia triads were reported, and there was absolutely no evidence of any demyelination seen in any of the brains autopsied [11]. If

there was brain hypoxia, it was of such short duration that not even demyelination from the hypoxia death of oligodendrocytes could occur [12]. Therefore, brain hypoxia is probably not the main explanation for the microglial nodules. However, there is another
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very well-documented explanation for the microglial nodules that were found in these brain autopsies.

Extensive Evidence of Microglial Brain Nodules in *T. gondii* Infections

T. gondii infections have shown an ability to use microglia as "Trojan Horses" for intracellular tachyzoite replication and spreading these active protozoan parasites throughout the brains of infected hosts [14]. Infected microglia once activated have demonstrated an ability to transmigrate through astrocytes, which are part of the brain parenchyma and BBB [14]. Rapid transfers of these pathogens from infected glial cells to effector T cells has also been demonstrated [14]. Microglial nodules in the brain and CNS have also been widely observed in AIDS patients, where the microglial nodules were caused by *T. gondii* infections, cytomegalovirus infections or HIV itself [14-16]. Activated microglia from reactivated *T. gondii* have also been observed to cause neuronal apoptosis, which could eventually lead to neuronophagia [16]. *T. gondii* activated microglia also lead to elevated levels of the inflammatory cytokines IL-1β, IL-6, and TNF-α [16].

In view of the preceding *T. gondii* symptoms, it is significant that microglial activation was also found in 81% of the brains of COVID-19 fatalities, and microglial nodules were found in 63% of the brains of COVID-19 fatalities [11]. But while neurons were found in some microglial nodules, no apoptotic neuron-astrocyte-microglia triads were reported, and there was absolutely no demylination found in any of the brains autopsied [11]. But one of the main consequences of brain hypoxia, BBB breakdown and oxidative and inflammatory stress is axonal demyelination of the neurons [12]. If there was brain hypoxia, it was of such short duration that not even demyelination occurred. Therefore, *T. gondii* infections are a more likely explanation for the microglial activation and microglial nodules found in the reported brain autopsies.

Conclusive evidence will only become available after a major research effort requiring major resources in equipment and money. Publication of this paper is needed first in order to muster the resources in equipment and money needed to collect the necessary conclusive evidence.

Reviewer: 2 Comments to the Author This brief paper provides an interesting hypothesis about some peculiar aspects of COVID-19 epidemiology and physiopathology, which so far haven't received any attention, despite the massive amount of publications on COVID-19 appeared in the last 12 months. I feel that this hypothesis is worth reading and may suggest a new avenue of research, so far missed. The author has well incorporated all the comments received so far. The paper is easy to read and might attract readers' attention. I have one MAJOR comment: to facilitate the flow of ideas and facilitate reading, the author should add several HEADINGS to split the manuscript in different sections. Although the manuscript taps on different issues, so far the paper doesn't include any headings, and this does not help the reader contextualize the paper content.

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The paper has been extensively rewritten, reorganized, and it now has several headings to

improve the clarity.

Reviewer: 3 Comments to the Author The paper develops an interesting topic, that currently has several questions and few answers. The hypothesis pointed out by the authors is very interesting and deserves attention for future researches in the literature. However I have some queries for the authors. From line 54 to line 67 authors cite results from the review from Xiao et al., 2018 claiming that " these neurotransmitter disruptions have been hypothesized to cause schizophrenia" However the review explains that the IgG levels could be also associated to the antipsychotic drug use, and authors should mention this in the text. Moreover the same review associates T.gondii IgG also to other psychiatric disorder and cognitive impairment. The authors should mention this in the text as well.

This is what Xiao et al. actually stated in their paper [10].

"It is also possible that the antiparasitic effects of some of the medications commonly used to treat schizophrenia affect the levels of antibodies. Consistent with this possibility is the recent report of increased levels of Toxoplasma antibodies in individuals with treatment resistant forms of schizophrenia.

Although most studies of Toxoplasma have examined associations with schizophrenia, increased rates of exposure to Toxoplasma have been found in a range of other psychiatric disorders including psychotic-like symptoms, bipolar disorder, self-directed violence and suicide attempts, general anxiety disorders, mixed anxiety and depressive disorder, obsessive-compulsive disorder, Autism, and depression during pregnancy."

The main focus of my paper is on schizophrenia cases caused by *Toxoplasma gondii*. The *T. gondii* antibody levels may indeed change because of the medications, but this is tangential, because the main point is that the antibodies indicate infection by *T. gondii*, regardless of the increased numbers of antibodies caused by medications.

At last, only 3/34 individuals with schizophrenia were positive for the T.Gondii IgG, and this could explain the multifactoral risk factors for schizophrenia. At page 4 (line 73) authors should mention also sedentary lifestyle as a risk factor for schizophrenia.

I never said that all schizophrenia cases were caused by *T. gondii*, it would only require a small percentage, because even a very small percentage would be enough to explain the higher mortality rate for schizophrenia patients infected by COVID-19.

A sedentary lifestyle may be a risk factor, but this is tangential to the main point about some schizophrenia cases being caused by *T. gondii*.

A better replacement paragraph is now used to summarize the schizophrenia discussion.

Increased COVID-19 Mortality of Patients With Schizophrenia

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A strong link between schizophrenia and a higher mortality rate from SARS-CoV-2 has been observed, with a mortality odds ratio (O.R.) of 2.67 above normal patients, after adjustment of the O.R. for age, sex, race and extra risk factors [4]. The extra schizophrenia risk factors are smoking, cardiovascular disease, diabetes, chronic respiratory disease, antipsychotic medication effects, etc. [5].

Still at page 4 (lines 90-92) authors affirm that "These higher mortality rates closely match corresponding increases in latent pathogen infection rates, particularly protozoan parasite infections, such as T. gondii. At the light of the current evidence authors should "soften" this sentence.

The paragraph has been modified. It now states -

There are significant matches between the mortality rate O.R. of these individual categories and their corresponding odds ratios of *T. gondii* infections [11-14]. In 6,663 German adults, the detection of serum IgG antibodies to *T. gondii* progressively increased from 20% in the 18 to 29 age group to 77% in the 70 to 79 age group [14]. Males had increased *T. gondii* infection rates compared to females (O.R. of 1.8), and there were increased *T. gondii* infection rates for individuals having body mass indexes over 30 (O.R. of 1.3) [14]. If *T. gondii* infection rates are similar among the U.S., the U.K. and Germany, these corresponding O.R. matches in specific categories of individuals between increased COVID-19 mortality rates and *T. gondii* infection rates suggest synergistic immune system dysfunctions, including CD8 T cell exhaustion, worsening COVID-19 and *T. gondii* infections. This synergy can explain several unusual COVID-19 characteristics. This synergy would also explain Long COVID-19.

From lines 163-171 authors mention several other pathogen synergies. In my opinion authors should move this paragraph at page 2 after line 37.

I believe that doing this would not help the flow of the discussion.

At the end of the paper, authors should make some recommendations for researchers like suggesting a T.Gondii IgG research in patients with long COVID 19 or case control studies in patients with or without neurological or psychiatric symptoms following the infection.

This is implemented. Recommendations for the researchers are provided in an added paragraph.

Suggestions for Researchers

One way to directly prove the connection between Long COVID-19 and toxoplasmosis would be to measure the IgG antibodies to *T. gondii* in the blood of people suffering from Long COVID-19, and compare the IgG antibody measurements to people who did not suffer any Long COVID-19. Other options would include taking CT scans of the brains

or brain tissue samples from both groups of people during postmortem autopsies to compare *T. gondii* infection rates to instances of Long COVID-19 or fatal COVID-19.

Reviewer: 4 Comments to the Author The submitted manuscript offers a hypothesis on how latent protozoan infections such as t. gondii may be responsible for differences in risk of death and risk of persistent symptoms due to Covid-19 in various populations. Latent infections as a risk factor for Covid-19 mortality: The author describes increased prevalence of T. gondii antibodies found in patients with certain psychiatric conditions, in men (compared to women), those in older age groups, and adults with increased BMI--groups that are at increased risk of Covid-related mortality. He argues that latent parasitic infections may be responsible for increased risk in these populations. While this may be possible, what is the evidence for causation?

Some stronger evidence is now included in the revised paper with the addition of several new references.

Evidence of Microglial Activation and Microglial Nodules in Brain Autopsies of COVID-19 Fatalities

It has been recently reported that 41 patients, with ages ranging from 38 to 97 years, who died from COVID-19, had brain autopsies in 2020 at the Columbia University Irving Medical Center [11]. The patients tested before death showed elevated inflammatory

cytokines, including elevated IL-6 in 26 (96%) of the 27 patients tested [11]. Neuropathological examination of 20 to 30 areas from each brain found microglial activation in 34 of the 41 brains (81%) [11]. Microglial clusters (microglial nodules) were found in 26 of the brains (63%), being most prevalent in the brainstem, and neurons were found in some of the microglial nodules, indicating neuronophagia [11]. However, viral RNA and viral proteins were not significantly detected in the brain cells or the microglial nodules, while acute and subacute hypoxic damage was seen in every brain; thus the formation of the microglial nodules was attributed to hypoxia, not SARS-CoV-2 [11].

It is well documented that brain hypoxia disrupts the BBB, causing leakage of proinflammatory plasma proteins, including immunoglobulins, fibrinogen and complement [12]. It is also well documented that microglial activation and perivascular microglial clusters can be caused by fibrin deposition from perivascular leakage of plasma protein fibrinogen into areas of BBB disruption [13]. In addition, the formation of apoptotic neuron-astrocyte-microglia triads and extensive damage to the myelin sheath (demyelination) from oxidative stress and inflammatory stress has been well documented for brain hypoxia/ischemia [12]. But while neurons were found in some microglial nodules, no apoptotic neuron-astrocyte-microglia triads were reported, and there was absolutely no evidence of any demyelination seen in any of the brains autopsied [11]. If there was brain hypoxia, it was of such short duration that not even demyelination from the hypoxia death of oligodendrocytes could occur [12]. Therefore, brain hypoxia is probably not the main explanation for the microglial nodules. However, there is another very well-documented explanation for the microglial nodules that were found in these brain autopsies.

Extensive Evidence of Microglial Brain Nodules in *T. gondii* Infections

12

T. gondii infections have shown an ability to use microglia as "Trojan Horses" for intracellular tachyzoite replication and spreading these active protozoan parasites throughout the brains of infected hosts [14]. Infected microglia once activated have demonstrated an ability to transmigrate through astrocytes, which are part of the brain parenchyma and BBB [14]. Rapid transfers of these pathogens from infected glial cells to effector T cells has also been demonstrated [14]. Microglial nodules in the brain and CNS have also been widely observed in AIDS patients, where the microglial nodules were caused by *T. gondii* infections, cytomegalovirus infections or HIV itself [14-16]. Activated microglia from reactivated *T. gondii* have also been observed to cause neuronal apoptosis, which could eventually lead to neuronophagia [16]. *T. gondii* activated microglia also lead to elevated levels of the inflammatory cytokines IL-1β, IL-6, and TNF-α [16].

In view of the preceding *T. gondii* symptoms, it is significant that microglial activation was also found in 81% of the brains of COVID-19 fatalities, and microglial nodules were found in 63% of the brains of COVID-19 fatalities [11]. But while neurons were found in some microglial nodules, no apoptotic neuron-astrocyte-microglia triads were reported, and there was absolutely no demylination found in any of the brains autopsied [11]. But one of the main consequences of brain hypoxia, BBB breakdown and oxidative and inflammatory stress is axonal demyelination of the neurons [12]. If there was brain hypoxia, it was of such short duration that not even demyelination occurred. Therefore, *T. gondii* infections are a more likely explanation for the microglial activation and

microglial nodules found in the reported brain autopsies.

Conclusive evidence will only become available after a major research effort requiring major resources in equipment and money. Publication of this paper is needed first in order to muster the resources in equipment and money needed to collect the necessary conclusive evidence. This paper is proposing an explanation. Other researchers with far more resources will be able to verify it or disprove it.

It seems plausible that an underlying immune mechanism may predispose people to both Covid-19 and risk of other infections, but it's not clear based on the presented evidence that 1) this mechanism is unique to latent protozoan infections or that 2) latent protozoan infections cause the immune disruption that leads to poor Covid-19 related outcomes. Support for the first argument may come from studies that suggest that these risk factors (male sex, severe mental illness, increased BMI, older age) are unique to Covid-19 and T. Gondii, as opposed to risk factors for worse infectious outcomes in general. Support for the second argument could come from studies suggesting that latent protozoan infections have an adverse impact on outcomes in the setting of subsequent infections. Is there evidence for the "bidirectional synergy" or "mutually beneficial immune cell dysfunctions" referenced? What are these mechanisms, specifically? What is meant by "beneficial"?

Beneficial means beneficial for the pathogens. Synergy between viral infections and latent protozoan infections is not rare. For example, there is a bidirectional synergy between HIV infections and protozoan parasite infections that enhance the ability of HIV 13

to enter CD4 T cells by the CCR5 receptor, and also helps the protozoan parasites. Protozoan parasites *Leishmania infantum* and *Leishmania donovani* can accelerate HIV infections by causing over-expression of CCR5, a co-receptor for HIV entry into CD4 T cells. This unusual but fundamental host immune system manipulation during such protozoan infections provides assistance to concurrent HIV infections.

See Lindoso, J.A.L., Moreira, C.H.V., Cunha, M.A., Queiroz, I.T. Visceral Leishmaniasis and HIV coinfection: current perspectives. *HIV AIDS(Auckl).* 2018;**10**:193-201

Latent infections as a risk factor for long COVID-19: The author discusses the possibility that a variety of latent infections (viral, bacterial, protozoan) can act in synergy and contribute to symptoms of post-acute Covid-19 (long Covid). Is there evidence that the same risk factors for Covid-19 mortality referenced earlier in the paper are risk factors for long-Covid?

Many of the strangest symptoms of Long COVID-19 are also seen in COVID-19. And these symptoms are either seen in toxoplasmosis, or made entirely possible by cranial nerve palsies caused by reactivated *T. gondii*. Also see the added paragraph with Suggestions for Researchers immediately below for suggestions on collecting direct evidence.

The author describes how symptoms of long-Covid parallel symptoms of toxoplasmosis reactivation; how would you distinguish between the two?

Many of the strangest symptoms of Long COVID-19 are also seen in toxoplasmosis. A paragraph is added to suggest ways to collect the evidence to clarify the relationship.

Suggestions for Researchers

One way to directly prove the connection between Long COVID-19 and toxoplasmosis

would be to measure the IgG antibodies to *T. gondii* in the blood of people suffering from Long COVID-19, and compare the IgG antibody measurements to people who did not suffer any Long COVID-19. Other options would include taking CT scans of the brains or brain tissue samples from both groups of people during postmortem autopsies to compare *T. gondii* infection rates to instances of Long COVID-19 or fatal COVID-19.

Summary: This paper presents an interesting hypothesis concerning the possibility that latent parasitic infections contribute to adverse outcomes in the acute and post-acute phase of Covid-19 infection. Though shared immune mechanisms may contribute to increased risk of Covid-19 infection and other infections, it isn't clear based on the evidence presented that latent infections are a cause of adverse outcomes across various at-risk groups. Suggestions regarding how this hypothesis could be tested may help guide future research in this area.

Actually, stronger evidence is now included in the revised paper with the addition of several new references.

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Evidence of Microglial Activation and Microglial Nodules in Brain Autopsies of COVID-19 Fatalities

It has been recently reported that 41 patients, with ages ranging from 38 to 97 years, who died from COVID-19, had brain autopsies in 2020 at the Columbia University Irving Medical Center [11]. The patients tested before death showed elevated inflammatory cytokines, including elevated IL-6 in 26 (96%) of the 27 patients tested [11]. Neuropathological examination of 20 to 30 areas from each brain found microglial activation in 34 of the 41 brains (81%) [11]. Microglial clusters (microglial nodules) were found in 26 of the brains (63%), being most prevalent in the brainstem, and neurons were found in some of the microglial nodules, indicating neuronophagia [11]. However, viral RNA and viral proteins were not significantly detected in the brain cells or the microglial nodules, while acute and subacute hypoxic damage was seen in every brain; thus the formation of the microglial nodules was attributed to hypoxia, not SARS-CoV-2 [11].

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Extensive Evidence of Microglial Brain Nodules in *T. gondii* Infections

T. gondii infections have shown an ability to use microglia as "Trojan Horses" for intracellular tachyzoite replication and spreading these active protozoan parasites

throughout the brains of infected hosts [14]. Infected microglia once activated have demonstrated an ability to transmigrate through astrocytes, which are part of the brain parenchyma and BBB [14]. Rapid transfers of these pathogens from infected glial cells to effector T cells has also been demonstrated [14]. Microglial nodules in the brain and CNS have also been widely observed in AIDS patients, where the microglial nodules were caused by *T. gondii* infections, cytomegalovirus infections or HIV itself [14-16]. Activated microglia from reactivated *T. gondii* have also been observed to cause neuronal apoptosis, which could eventually lead to neuronophagia [16]. *T. gondii* activated microglia also lead to elevated levels of the inflammatory cytokines IL-1β, IL-6, and TNF-α [16].

In view of the preceding *T. gondii* symptoms, it is significant that microglial activation was also found in 81% of the brains of COVID-19 fatalities, and microglial nodules were

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found in 63% of the brains of COVID-19 fatalities [11]. But while neurons were found in some microglial nodules, no apoptotic neuron-astrocyte-microglia triads were reported, and there was absolutely no demylination found in any of the brains autopsied [11]. But one of the main consequences of brain hypoxia, BBB breakdown and oxidative and inflammatory stress is axonal demyelination of the neurons [12]. If there was brain hypoxia, it was of such short duration that not even demyelination occurred. Therefore, *T. gondii* infections are a more likely explanation for the microglial activation and microglial nodules found in the reported brain autopsies.

There are ways to test the relationship between the latent infections and the COVID-19 patients during and after their viral infection. And a paragraph is added to suggest ways to collect the evidence to clarify the relationship.

Suggestions for Researchers

One way to directly prove the connection between Long COVID-19 and toxoplasmosis would be to measure the IgG antibodies to *T. gondii* in the blood of people suffering from Long COVID-19, and compare the IgG antibody measurements to people who did not suffer any Long COVID-19. Other options would include taking CT scans of the brains or brain tissue samples from both groups of people during postmortem autopsies to compare *T. gondii* infection rates to instances of Long COVID-19 or fatal COVID-19.

3rd Editorial Decision

Decision Letter

Dear Dr Roe:

Thank you for revising and resubmitting your manuscript to the Journal of Neuroscience Research. We've now received the reviewer feedback and have appended those reviews below. There still some points that require your attention, but, they should all be relatively straightforward to address. If there are any questions or points that are problematic, please feel free to contact me.

We ask that you return your manuscript within 30 days. Please explain in your cover letter how you have changed the present version and submit a point-by-point response to the editors' and reviewers' comments. If you require longer than 30 days to make the revisions, please contact Dr Cristina Ghiani (cghiani@mednet.ucla.edu). To submit your revised manuscript: Log in by clicking on the link below https://wiley.atyponrex.com/submissionBoard/1/4b787f63-82b9-4300-ab0b-eb4237708b4c/current

(If the above link space is blank, it is because you submitted your original manuscript through our old submission site. Therefore, to return your revision, please go to our new submission site here (submission.wiley.com/jnr) and submit your revision as a new manuscript; answer yes to the question "Are you returning a revision for a manuscript originally submitted to our former submission site (ScholarOne

Manuscripts)? If you indicate yes, please enter your original manuscript's Manuscript ID number in the space below" and including your original submission's Manuscript ID number (jnr-2021-Feb-9560.R2) where indicated. This will help us to link your revision to your original submission.)

The journal has adopted the "Expects Data" data sharing policy, which states that all original articles and reviews must include a Data Availability Statement (DAS). Please see https://authorservices.wiley.com/author-resources/Journal-Authors/open-access/data-sharing-citation/data-sharing-policy.html#standardtemplates for examples of an appropriate DAS. Please include the DAS in the manuscript as well.

Thank you again for your submission to the Journal of Neuroscience Research; we look forward to reading your revised manuscript.

Best Wishes,

Dr Karina Alvina
Associate Editor, Journal of Neuroscience Research

Dr Cristina Ghiani
Editor-in-Chief, Journal of Neuroscience Research

EiC comments:
Two reviewers are overall enthusiastic and supportive of your manuscript, however, the third reviewer has still some concerns, which should be addressed before the paper is suitable for publication. The reviewers find the ideas in the paper to be provocative and important, however, due to the weakness in the evidence indicating causation, I suggest that the author tones down and softens his conclusions and speculations, and please takes in account the reviewer points.
Limitations exist and should be recognised and mentioned, as these, after all, may help to move forward the field.

In addition, the manuscript needs to be formatted using the guidelines to the authors:
https://onlinelibrary.wiley.com/page/journal/10974547/homepage/forauthors.html

Few points:
Please remove the list of abbreviations and, make sure all the abbreviations are spelled out the first time they are used in both the abstract and the main text.
JNR does not use the number-format for the references, and these should be listed in alphabetical order.
PLease

Associate Editor: Alvina, Karina
Comments to the Author:
Thank you for taking care of most of the reviewers' concerns, however one of the reviewer still raises important questions that need to be addressed properly.

Reviewer: 2

Comments to the Author
The author has made extensive revisions to the manuscript, which now sounds fine and well balanced. The paper should be published, because it includes some interesting insights, which may stimulate new avenues of research.

Reviewer: 3

Comments to the Author
The current version is significantly improved. The author answered to my queries in a satisfactory way

Reviewer: 1

Comments to the Author
Although the author provided additional arguments and references in an attempt to support his hypothesis on the involvement of Toxoplasma gondii on the increased mortality observed in individuals with low T cell numbers and COVID-19, this reviewer still believes that there is little evidence linking this protozoa and

SARS-CoV-2 infection. In fact, COVID-19 mortality has been shown to increase in individuals with fungal infections such as mucormycosis and aspergillosis. However, there is no one single case reported indicating the involvement of T. gondii in the mortality observed in patients with COVID-19. A simple explanation in older patient's susceptibility to this viral infection would be that as we age our thymus becomes smaller reducing T cell numbers and making us susceptible to infections. Mortality is also exacerbated by the fact that older individuals usually suffer of additional comorbidities. In summary, the idea that T. gondii might be responsible for the high mortality observed in COVID-19 patients is provocative due to the pathogenesis of the parasite and its CNS involvement but the arguments provided are weak to establish causation.

Authors' Response

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Response to the Reviewers' comments on the manuscript entitled *"An explanation for the higher mortality rate of COVID-19 patients having schizophrenia, the higher mortality rates of patients in other categories and for Long COVID-19"* submitted to the Journal of Neuroscience Research - Manuscript # jnr-2021-Feb-9560

Thank you very much for giving me the opportunity to amend the paper in response to the reviewers comments.

Response to All Comments - The reviewers have clearly spent time to read the paper, and these are very helpful comments, which are both valuable and sincerely appreciated. I have revised the paper to address the reviewers' comments and also have made wording revisions to improve the paper's clarity. I have indicated new text by red coloring. If I have missed anything, please let me know. I will address any problem.

Each Review Comment is listed in bold italics. The Responses and the Revisions are included just below each Reviewer Comment in RED. Revisions in paper are summarized below.

EiC comments: Two reviewers are overall enthusiastic and supportive of your manuscript, however, the third reviewer has still some concerns, which should be addressed before the paper is suitable for publication. The reviewers find the ideas in the paper to be provocative and important, however, due to the weakness in the evidence indicating causation, I suggest that the author tones down and softens his conclusions and speculations, and please takes in account the reviewer points. Limitations exist and should be recognised and mentioned, as these, after all, may help to move forward the field.

A very good point and I fully agree. The title and the tone of the entire paper have been substantially softened in numerous changes in the text. The title has also been softened - it is now:

A role for T cell exhaustion in Long COVID-19 and severe outcomes for several categories of COVID-19 patients

In addition, the manuscript needs to be formatted using the guidelines to the authors: https://onlinelibrary.wiley.com/page/journal/10974547/homepage/forauthors.html Few points: Please remove the list of abbreviations and, make sure all the abbreviations are spelled out the first time they are used in both the abstract and the main text. JNR does not use the number-format for the references, and these should be listed in alphabetical order.

I have removed the list of abbreviations, and the abbreviations are spelled out the first time they are used in both the abstract and main text. The references are now listed in

alphabetical order according to the name-year system.
2
Associate Editor: Alvina, Karina Comments to the Author: Thank you for taking care of most of the reviewers' concerns, however one of the reviewer still raises important questions that need to be addressed properly. Reviewer: 2 Comments to the Author The author has made extensive revisions to the manuscript, which now sounds fine and well balanced. The paper should be published, because it includes some interesting insights, which may stimulate new avenues of research. Reviewer: 3 Comments to the Author The current version is significantly improved. The author answered to my queries in a satisfactory way
Glad to hear it. Thank you.
Reviewer: 1 Comments to the Author Although the author provided additional arguments and references in an attempt to support his hypothesis on the involvement of Toxoplasma gondii on the increased mortality observed in individuals with low T cell numbers and COVID-19, this reviewer still believes that there is little evidence linking this protozoa and SARS-CoV-2 infection. In fact, COVID-19 mortality has been shown to increase in individuals with fungal infections such as mucormycosis and aspergillosis. However, there is no one single case reported indicating the involvement of T. gondii in the mortality observed in patients with COVID-19.
There is little evidence, because apparently nobody has thought of the possibility before. In fact, although this paper should still be valid and applicable to Long COVID-19 at the very least, the editors may decide not to publish this paper, because an Egyptian paper published a few weeks ago discussed a clinical study that concluded that protozoan parasite infections can actually moderate COVID-19 viral infections by stimulating the release of higher levels of interferon-γ from T cells. This would be true - antiviral T cell functions could operate normally and release interferon-γ to suppress viral infections, IF there is no T cell exhaustion because of early drug treatments specifically for the protozoan parasite infections, or because of other factors that avoided T cell exhaustion. I have added several paragraphs to clarify the possibilities and explain the circumstances where T cell exhaustion is blocked and normal antiviral T cell functions could apply. I also point out there are at least 11 different genetic types of *T. gondii* with vast differences in their immunological consequences.
However, the journal editors may decide not to publish this paper because of this Egyptian clinical study paper. I would understand such a decision.
There Are Several Types of *T. gondii* With Very Substantial Differences
It is important to note that there are actually at least 11 different genetic types (haplogroups) of *T. gondii*, with major differences in their growth, migration and transmission characteristics, and very substantial differences exceeding five orders of magnitude in terms of their effects on the immune system and their virulence (Halonen et al., 2013). It should also be noted that there is a very significant geographical influence on the distribution of the specific *T. gondii* haplogroups (Halonen et al., 2013). For example, the *T. gondii* haplogroups
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(predominantly Type II) of North America are the same as in Europe, but they are much less virulent compared to the very virulent *T. gondii* haplogroups (e.g., Type I) of South America (Halonen et al., 2013). Therefore, the specific

haplogroup of *T. gondii* being studied in a specific continent should be identified and specified because of their very substantial immunological differences.

Early Drug Treatments for Toxoplasmosis Can Block T Cell Dysfunctions and T Cell Exhaustion, and Could Maintain Effective T Cell Functions to Block Latent Pathogen Reactivations and Moderate Later Viral Infections

There are several drug treatments for *T. gondii* infections, such as sulfamethoxazole and trimethoprim, and it has been demonstrated that early initiation of such drug treatments for *T. gondii* infections can greatly reduce later induced T cell dysfunctions, including CD8 T cell exhaustion, in the late stage of chronic infection (Bhadra et al., 2011). CD8 T cells normally produce interferon-γ and cytotoxic granzyme B to control toxoplasmosis and block reactivation of *T. gondii* cysts, but these are also generally used to suppress viral infections (Bhadra et al., 2011). Therefore, if T cell exhaustion and other dysfunctions can be blocked by early drug treatments for protozoan parasite infections, then effective CD8 T cell responses to later viral infections, including SARS-CoV-2, could essentially be maintained (Bhadra et al., 2011). These drug treatments, if timely, could essentially mitigate the increased risk of mortality of COVID-19 patients having protozoan parasite infections.

In the Absence of T Cell Exhaustion, Latent Pathogen Infections Can Sometimes Improve Immune Responses to Other Pathogens

In summary, T cell exhaustion is caused by the combination of long duration pathogen infections or cancers that produce inflammation and long duration antigen stimulation with significant titers of antigens. However, if one or more of these criteria is not satisfied, it is possible for a relatively recent latent pathogen infection, or a latent pathogen infection producing little inflammation or a low titer of antigens, to induce very little or no T cell exhaustion. Furthermore, if some circumstance can block T cell exhaustion that reduces T cell functions, some latent pathogen infections, including some types of protozoan parasite infections, can induce CD4 T cells and CD8 T cells to release higher levels of antiviral interferon-γ that can promote an improved response to a later infection by a virus (Abdel-Hamed et al., 2021; Bhadra et al., 2011).

A recent paper summarized a clinical study of 375 COVID-19 patients in Egypt and the relationship between their SARS-CoV-2 viral infection outcomes and the presence in almost 69% of the patients of various protozoan parasite infections, including *T. gondii* (Abdel-Hamed et al., 2021). However, there was no indication of whether or not any of these patients had received early drug treatments for their protozoan parasite infections that would have blocked later T cell exhaustion (Abdel-Hamed et al., 2021). This clinical study concluded that the protozoan parasite stimulation of T cells significantly increased the levels of interferon-γ, and that existing protozoan parasite infections with increased levels of interferon-γ resulted in a large majority of cases with more moderate COVID-19

symptoms (Abdel-Hamed et al., 2021). It should be noted that this study did not address the following: (1) whether or not the protozoan parasite infected patients had previously received any early anti-protozoan parasite drug treatments that would have blocked T cell exhaustion and dysfunctions, thereby improving later T cell responses to COVID-19, (2) the vastly different immune effects of the various haplogroups of *T. gondii*, (3) the possibility or consequences of T cell exhaustion from protozoan parasite infections that

satisfied the necessary criteria to induce T cell exhaustion, (4) an opposite role for SARSCoV-2 in reactivating latent infections of *T. gondii* and (5) the possibility that such latent pathogen reactivations could produce the symptoms now called Long COVID-19 (Bhadra et al., 2011).

A simple explanation in older patient's susceptibility to this viral infection would be that as we age our thymus becomes smaller reducing T cell numbers and making us susceptible to infections. Mortality is also exacerbated by the fact that older individuals usually suffer of additional comorbidities. In summary, the idea that T. gondii might be responsible for the high mortality observed in COVID-19 patients is provocative due to the pathogenesis of the parasite and its CNS involvement but the arguments provided are weak to establish causation.

It is very true that the thymus disappears with age, and this certainly does not help the adaptive immune system of older people adapt to new pathogens. And it is also true they suffer additional comorbidities as they age. I have modified the end of the paragraph to briefly list these other effects of old age.

....Therefore, older individuals will generally also be more likely to have more severe T cell exhaustion with all its consequences for higher mortality. This would be yet another logical explanation for the increasing mortality rate of older individuals infected by SARS-CoV-2, in addition to the increasing number of comorbidities and general weakening of the immune system with increasing age.

There is little evidence, because apparently nobody has thought of the possibility before. Beside the very extensive evidence of T cell exhaustion and increased mortality for cancer patients and some pathogen infections including *T. gondii*, the evidence regarding its application to COVID-19 is sparse. If this paper is ever published, it might motivate other researchers, who have the necessary resources in money, equipment and manpower, to either prove or disprove this paper.

4th editorial decision

Decision Letter

Dear Dr Roe:

We thank the author for taking the reviewer's comments into consideration and making the requested changes. The manuscript is now suitable for publication. Thank you for submitting your work to our journal.

In the coming weeks, the Production Department will contact you regarding a copyright transfer agreement and they will then send an electronic proof file of your article to you for your review and approval.

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Congratulations, and thank you for choosing the Journal of Neuroscience Research for publishing your work. I hope you will consider us for the publication of your future manuscripts.

JOURNAL OF Neuroscience Research

Sincerely,

Dr Karina Alvina
Associate Editor, Journal of Neuroscience Research

Dr Cristina Ghiani
Editor-in-Chief, Journal of Neuroscience Research

Author response